Field Evaluation of Different Pesticides against Cotton Bollworms and Sucking Insects and Their Side Effects

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Abstract: Field experiments were conducted during 2010 and 2011 cotton growing seasons at Sakha Agricultural Research Station Farm to evaluate the efficacy of five insecticides, i.e., two synthetic pyrethroids (∝-cypermethrin, lambda-cyhalothrin), two organophosphorus (profenophos, chlropyrifos) and one carbamate (methomyl), against both pink bollworm (PBW), *pectinophora gossypiella* (saund) and spiny bollworm (SPW), *Earias insulana (Boisd)* and their effects against sucking insects; cotton aphid, *Aphis gossypii* (Golv), whitefly, *Bemisia tabaci* (Genn.) and Jassid, *Empoasca spp.* and their associated natural enemies,(*chrysopa sp., Paederus alfierii, Orius spp, Scymnus spp. and* True spider). Biochemical studies on bollworms larvae were conducted as well. The obtained results indicated that the tested synthetic pyrethroids were the most efficient compounds during the two seasons. The treatments could be arranged descendingly according to the average of the two seasons as follows; ∝-cypermethrin (81.45%), lambda-cyhalothrin (71.91%), methomyl (68.33%), profenophos (66.75%) and chlorpyrifos (62.58%) against PBW; and were ∝-cypermethrin (83.00%), lambda-cyhalothrin (81.61%), methomyl (81.15%), profenophos (78.87%) and chlorpyrifs (70.05%) against SPW. Regarding sucking insects, ∝-cypermethrin was efficient against aphid followed by profenophos, lambda-cyhalothrin, chlorpyrifos and methomyl meanwhile in case of whitefly (mature and immature stages) and Jassid all the tested insecticides induced a weak to moderate effect. In addition, ∝-cypermethrin, lambda-cyhalothrin and profenophos were more toxic against predators than chlorpyrifos and methomyl which induced a moderate effect. As for biochemical assays in PBW larvae, data indicated that both chlorpyrifos and profenophos-treated strains, expressed higher levels of Acetylcholinesterase (AChE) activity than the reference (Lab-susceptible) strain. As respects SPW larvae, data showed that chlorpyrifos-treated strains expressed higher levels of AChE activity than the reference (Lab- strain). Data also revealed that, relatively higher activity of glutathione-S-transferase (GST) has been observed in chlorpyrifos and profenophos- treated strains, over that of the lab-strain of PBW larvae. The same trend of data was obtained for SPW larvae.

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1. Introduction

In Egypt, cotton is one of the most important cash crops and represents more than half the income of two million small-scale farmers. But cotton is attacked by many insect species. Cotton bollworms are the most destructive pests infesting cotton plants. Pink bollworm (PBW), *pectinophora gossypiella* (saund) and spiny bollworm (SPW), *Earias insulana (Boisd)* infest many cotton producing areas of the world and cause a severe reduction in cotton yield and quality (Lohag and Nahyoon, 1995). Chemical control is still adopted as one of the major techniques for combating these serious pests.

The effectiveness of different pesticides against bollworms was studied by several authors (Khan *et al.* 2007; Balakrishman *et al*. 2009 and Magdy *et al.* 2009). Besides, the two pests, sucking insect pests; cotton aphid, whitefly, and Jassid attack cotton plants at different growing stages. Heavy infestation with these sucking insects causes extensive reduction in cotton yield and quality (Harris *et al*. 1992). The side effect of insecticides was evaluated against their associated natural enemies (Balakrishman *et al*. 2009).

However, the achieved control is not successful enough because of the insect's high capacity to develop resistance toward the majority of these compounds. AChE is a key enzyme in the insect nervous system that hydrolyzes acetylcholine neurotransmitter to terminate nerve impulses, and it is the primary target of organophosphate (OP) and carbamate insecticides. Insects have developed resistance to OPs and carbamates through modification of their AChE sensitivity to insecticides (Russell *et al*. 2004). The insensitive AChE in resistant strains sometimes, but not always, shows a reduced activity to substrates (Tan *et al*. 2008). The faster degradation of insecticides by metabolic enzymes is one such mechanism commonly associated with insecticide resistance. The involvement of GST, carboxylesterase, and microsomal monooxygenase in insecticide resistance has been reported in insecticide-resistant strains of many insect species (Sun, 1992).

The present study was conducted to evaluate the effect of some insecticides against pink bollworm, spiny bollworm, sucking insects (i.e., cotton aphid, whitefly, and jassid) and their side effects against some associated natural enemies under field conditions. Also some biochemical studies were carried out on the larvae of bollworms collected from insecticides-treated field to determine the activities of AChE and GST.

2. Materials and methods

2.1. Pesticides used:

Commercial formulations of five insecticides were used in the present study. Such insecticides were ∝-cypermethrin (Alphazid EC 10%) obtained from Kafr EL-Zayat Pesticides and Chemicals Co., applied at rate of 250 cm^3/fed; Lambda-cyhalothrin (Karate EC., 2.5 %) obtained from Samtrade Co., applied at rate of 750 cm^3/fed; Profenophos (Selecron EC., 72 %) obtained from Kafr EL-Zayat Pesticides and Chemicals Co., applied at rate of 750 cm^3/fed; Chlorpyrifos (Dursban EC., 48 %) obtained from Agrochem. Co., applied at rate of 1 L/fed. Methomyl (Lannate SP., 90 %) obtained from Kafr EL-Zayat Pesticides and Chemicals Co., applied at rate of 300 gm/fed.

2.2. Field experiment:

Experiments were conducted during 2010 and 2011 cotton growing seasons at Sakha Agricultural Research Station Farm. The cultivated cotton variety was Giza 89. Treatments were distributed in a complete randomized block design with four replicates for each treatment. The area of each replicate was one kirate (1/24 feddan, 175m^2) and four kirate were used as untreated control. All agricultural processes were carried out as usual.

Each of the tested insecticides was applied three times at two weeks intervals in between. The insecticides were diluted with water 200 L/ fed. and sprayed using a knapsack sprayer with one nozzle (Mod Cp3). The first spray took place on 18/7/2010 and 21/7/2011 for pyrethroids and (O.P. and carbamate) in the two seasons, respectively.

Samples of 100 green bolls per treatment (25 bolls for each replicate) were taken at random and dissected. Percents of infestation were estimated immediately before the first spray and then every week through out the period of experiment. Henderson and Tilton equation (1955) was used to calculate the reduction percentage of infestation.

Regarding the sucking insects; aphid, whitefly and Jassid, 25 cotton leaves per replicate were collected randomly from bottom, middle and the top of the cotton plant (2+1+2 leaves per plant, respectively). The upper and lower leaf surfaces were inspected in the field using suitable lens to count the number of immature stages of whitefly. Sampling and counting were made before the first spray and 7 and 14 days after each spray. Associated natural enemies (*Chrysopa sp paederus alfierii, Orius spp. Scymnus spp.* and True spider) were also counted on 100 cotton plants for each treatment. Reduction percentages were calculated according to Henderson and Tilton equation 1955.

2.3. Biochemical studies:

2.3.1. Strains:

Two different strains of PBW and SBW were used, these strains were: a- The Laboratory strain was obtained from the Bollworms Research Department, Plant Protection Research Institute, Dokki, Giza, Egypt. b- The larvae of field strains were collected before and after spraying of different insecticides.

2.3.2. Sample preparations:

Ten larvae from each strain were weighted and homogenized with cold phosphate buffer PH 7.2 (2 gm larvae/10 ml phosphate buffer) using a glass homogenizer. The cold crude extracts were centrifuged at 4000 rpm for 20 minutes using a cooling centrifuge, and then passed through glass wool to remove the last of insoluble cell debris and lipids. The supernatant was kept in deep freezer at -20^{o}C until it was used for the determination of total protein, AChE and GST assays. Total protein concentration, AChE and GST activity were determined according to the method of Lowry *et al* (1951), kinetic method of Den Blawen *et al*. (1983) and method of Habig *et al*. (1974), respectively.

2.4. Statistical analysis:

Statistical analysis of data was carried out according to Duncan multiple range test (Duncan, 1955).

3. Results and Discussion

3.1. The effect of the tested compounds against cotton bollworms:

The efficiency of five insecticides belonging to three different chemical group, pyrethroids, O.P and carbamate on the bollworms which infested cotton plant was evaluated in a field trial during 2010 and 2011 seasons. The number of the larvae of pink and spiny bollworms in the green bolls was recorded before and after treatment and the percent of reduction was calculated (Table 1, 2).

The obtained results in Table (1) indicated that, the toxic effect of the tested compounds against PBW in 2010 and 2011 cotton seasons, when they were applied once, twice and triple. Based on the general mean of reduction percentage in infestation of PBW in 2010 season, it was ranged between 63.03 to 81.96 %. In this respect the tested pyrethroids, ∝-cypermethrin and lambda-cyhalothrin were the most effective compounds as they caused 81.96 and 74.78% reduction respectively, followed by methomyl causing 68.92% reduction. While the two tested O.P. insecticides profenophos and chlorpyrifos were the least effective recording 67.76 and 63.03%, respectively. In 2011, the percentage of reduction in PBW larval population was lesser than in 2010 season and could be arranged descendingly as follows: ∝-cypermethrinn (80.93%), Lambda-cyhalothrin (69.04%), methomyl (67.84%), profenophos (65.73%) and chlorpyrifos (62.17%). The same trend of data was obtained for SPW larvae (Table 2).

Insecticide efficacy depends on the initial activity of the active ingredient on the target pest and its residual activity (persistence), which are both influenced by environmental parameters such as temperature, sunlight, or rainfall (Mulrooney and Elmore, 2000). The efficiency of the tested pyrethroids depend upon the active chemical groups in each pyrethroid, nature and ratio of optical and geometric isomers which allow a certain degree of effectiveness, the physical properties which determine the degree of penetration and volatility which can increase or decrease the efficiency (Abo-Sholoa *et al*. 2000). In this respect, the toxicity of the different pyrethroids on the field strain of pink and spiny bollworms could be demonstrated according to the differences in their chemical structure. Perusal of these results clearly exhibited that, all the recorded percentages of reduction in growing season 2010 were higher than those obtained in the growing season 2011. The obtained percentage of reduction ranged from 77.65 to 83.31 comparing with 62.46 to 82.69 with the latter season. The results obtained at season 2010 (Table 1 and 2) indicated that, the tested pyrethroid insecticides (∝-cypermethrin and Lambda-cyhalothrin) were more efficient in controlling the bollworms larval population than the used carabamate and O.P. compounds, which could be related to the reduction in their use in this area (Ishtiaq *et al*. 2012).

These results agree with those obtained by Khattak *et al*. (2004), khan *et al*. (2007) and Balakrishman *et al*. (2009), who indicated that Karate 2.5 EC (lamdba-cyhalothrin), Sherpa 5% EC (cypermethrin) and bifenthrin 10 EC at their recommended rates were more effective in reducing the incidence of bollworms. El-Basyoui (2003) found that synthetic pyrethroids were the most efficient compounds compared with O.P and carbamate insecticides of the larvae of bolloworms. Also Younis *et al*. (2007) reported that, the synthetic pyrethroid, Lambda-cyhalothrin and deltamethrin exhibited the greatest reduction in bollworms infestation compared to the organophosphrus pesticide treatment (chlorpyrifos and profenofos). On the other hand, Mahar *et al*. (2004) found that chlorpyrifos was effective on PBW.

3.2. The effect of the tested compounds against aphid, whitefly and Jassid infestation:

Based on the average of reduction percentage of the two seasons, the effectiveness of the tested compounds against sucking insects was shown in tables (3 to 6). Data presented in Table (3) summarize the toxic effect of the five compounds against aphid in 2010 and 2011 cotton seasons, the obtained results revealed that, ∝-cypermethrin was effective than the other tested compounds as it caused 78.1% reduction in infestation, followed by profenophos, lambda-cyhalothrin and chlorpyrifos since they caused 74.8, 74.1 and 73.4% reduction in infestation, while methomyl had a moderate effect, where it gave 68.9% reduction.

In case of whitefly (immature stages), data presented in Table (4) revealed that all the tested compounds exhibited a moderate reduction in infestation, ∝-cypermethrin was the most effective recording 47.1% reduction in infestation followed by chlorpyrifos (40.5%), lambda-cyhalothrin (39.7%), profenophos (39.5%) and methomyl (38.2%). Concerning whitefly (mature stages), data presented in Table (5) showed that, profenophos had the highest effect recording 54.7% reduction followed by chlorpyrifos (44.5%), methomyl (43.3%), lambda-cyhalothrin (41.7%) and alpha-cypermethrin (40.04%).

With regard to Jassid, data presented in Table (6) revealed that, chlorpyrifos was the most effective pesticide causing 59.9% reduction in infestation followed by profenophos (56.6%), lambda-cyhalothrin (55.4%) and alpha-cypermethrin (54.7%), while methomyl was the least effective recording 52.2% reduction.

These results were in a good harmony with those obtained by Abdel-Rahman *et al*. (1998) who found that Decis, Selecron and lannate showed strong pronounced effects on aphid, whitefly and Jassid. Asi *et al*. (2008) reported that, monocrotophos and endosulfan caused significant mortality of whitefly and Jassid until 168 hours after spray. Shawir *et al*. (2002) reported that profenofos, methomyl and deltamethrin were the most promising insecticides for controlling whitefly. Muthukumar and kalyanasundaram (2003) found that, the reduction of whitefly population was the highest with trizophos and profenofos followed by diflubenzuron, lambda-cyhalothrin and phosalone. In contrast to our findings, El-Zahi and Arif (2011) found that alpha-cypermethrin and Lambda-cyhalothrin failed to introduce sufficient control against aphids where they showed very feeble mean of effect recording 19.22% and 46.98% reduction, respectively.

Table (1): The insecticidal efficiency of the tested insecticides, against pink bollworm, *pectinophora gossypiella* (saund.) during 2010 and 2011 cotton seasons

Treatments	Rate of application	Seasons	Pre-spray	Number of larvae/ 100 bolls: 1st spray, Week after, 1 W	1st spray, 2 W	2nd spray, Week after, 1 W	2nd spray, 2 W	3rd spray, Week after, 1 W	3rd spray, 2 W	General mean	% Reduction: 1st spray, Week after, 1 W	1st spray, 2 W	2nd spray, Week after, 1 W	2nd spray, 2 W	3rd spray, Week after, 1 W	3rd spray, 2 W	General mean	Average of two seasons
Alpha-cyper methrin	250 Cm ^{3}/F	2010	3	2	1	5	3	2	4	3.33	75	88.89	72.2	85	84	86.66	81.96	81.45 ^{a}
		2011	3	1	2	3	4	6	5	4.33	83.33	77.78	84.62	80.96	76.48	82.45	80.93	
Lambda-cyhalothrin	750 Cm ^{3}/F	2010	2	2	2	3	3	3	3	3.00	62.5	66.67	75	77.5	82	85	74.78	71.91 ^{ab}
		2011	2	1	2	4	5	5	6	4.16	75.0	66.67	69.24	64.3	70.6	68.41	69.04	
Profenophos	750 Cm ^{3}/F	2010	3	3	3	7	6	7	9	6.33	62.5	66.67	61.11	70.0	72.0	70	67.76	66.75 ^{b}
		2011	3	2	4	6	8	7	9	6.50	66.66	55.56	69.24	61.92	72.56	68.44	65.73	
Chlorpyrifos	1000 Cm ^{3}/F	2010	2	2	3	4	4	6	7	4.66	62.5	50	66.68	70.0	64.00	65.0	63.03	62.58 ^{b}
		2011	2	2	2	4	5	6	8	4.83	50	66.67	69.24	64.3	64.72	57.88	62.17	
Methomyl	300gm/F	2010	2	2	3	3	4	4	4	3.66	62.5	50	75.01	70	76.0	80.0	68.92	68.33 ^{b}
		2011	3	2	4	5	6	5	9	5.66	66.66	55.56	74.36	71.44	70.6	68.41	67.84	
Untreated		2010	3	8	9	18	20	25	30	18.83	–	–	–	–	–	–	–	–
		2011	2	4	6	13	14	17	19	12.5	–	–	–	–	–	–	–	–

Values followed by different letters are significantly different.

Table (2): The insecticidal efficiency of the tested insecticides against spiny bollworm *Earias insulana* (Boised) during 2010 and 2011 cotton seasons

Treatments	Rate of application	Seasons	Pre-spray	Number of larvae/ 100 bolls: 1st spray, Week after, 1 W	1st spray, 2 W	2nd spray, Week after, 1 W	2nd spray, 2 W	3rd spray, Week after, 1 W	3rd spray, 2 W	General mean	% Reduction: 1st spray, Week after, 1 W	1st spray, 2 W	2nd spray, Week after, 1 W	2nd spray, 2 W	3rd spray, Week after, 1 W	3rd spray, 2 W	General mean	Average of two seasons
Alpha-cyper methrin	250 Cm ^{3}/F	2010	3	2	1	2	3	1	1	1.66	83.33	78.58	83.33	84.62	86.66	83.33	83.31	83.00 ^{a}
		2011	5	2	2	3	3	5	4	3.16	80	81.18	83.33	85	80	86.67	82.69	
Lambda-cyhalothrin	750 Cm ^{3}/F	2010	2	1	3	1	2	1	1	1.5	87.5	78.58	87.5	84.62	80	75	82.2	81.61 ^{b}
		2011	3	1	2	3	1	2	2	1.83	83.33	63.64	72.22	91.66	86.66	88.89	81.01	
Profenohpos	750 Cm ^{3}/F	2010	3	3	5	3	4	1	1	2.83	75	76.2	75	79.49	86.66	83.33	79.28	78.87 ^{c}
		2011	2	1	1	2	1	2	2	1.5	75	72.73	72.23	87.5	80	83.34	78.46	
Chlorpyrifos	1000 Cm ^{3} /F	2010	2	2	4	1	3	1	1	2.0	75	71.44	87.5	76.93	80	75	77.65	70.05 ^{d}
		2011	4	3	3	5	7	7	8	5.5	62.5	59.09	65.28	56.25	65	66.68	62.46	
Methomyl	300gm/F	2010	3	2	4	2	3	2	1	2.33	83.33	80.96	83.33	84.62	73.33	83.33	81.51	81.15 ^{b}
	300gm/F	2011	2	1	1	1	1	2	1	1.16	75	72.73	86.12	87.5	80	83.33	80.78	
Untreated		2010	8	14	8	8	13	5	4	8.66	–	–	–	–	–	–	–	–
		2011	5	10	11	18	20	25	30	19.0	–	–	–	–	–	–	–	–

Values followed by different letters are significantly different.

3.3. The side effect of the tested compounds:

3.3.1. The effect on the predators:

With regard to the associated natural enemies, (*Chrysopa sp., Paederus alfierii, Orius spp., Scymnus spp. and* True spider, data presented in table (7) based on the average of reduction percentage of the two seasons indicated that, all the tested compounds were toxic against the predators, the treatments could be arranged descendingly as follows: ∝-cypermethrin (73.5%), lambda-cyhalothrin (73.3%), profenophos (70.5%), chlorpyrifos (63.6%) and methomyl (60.2%). These results agree with the findings of Abdel-Rahman *et al*. (1998) who reported that, Selecron, Lannate and Decis exhibited a strong pronounced effects on associated predatory insects, (*Chrysoba spp., Conccinella spp. and paederus alferii*. Younis *et al*. (2007) revealed that, the synthetic pyrethroids, lambda-cyhalothrin, esfenvalerate and deltamethrin were the greatest in the reduction of the predator's population. Also El-Zahi and Arif (2011) showed that, chlorpyrifos, profenophos and methomyl proved to be the most toxic on associated predators.

Generally, it could be concluded that, two tested synthetic pyrethroids can be used to control bollworms in cotton fields, followed by carbamate and organophosphorus compounds. Regarding the examined sucking insects and their natural enemies, ∝-cypermethrin, lambda-cyhalothrin and profenophos only had a side effect in reducing the number of aphid and associated natural enemies.

3.3.2. The effect on cotton bollworms enzymes:

Biochemical assays in PBW larvae indicated that both chlorpyrifos and profenophos- treated strains, expressed higher levels of AChE activity than the reference (Lab- strain), representing 5.897 and 5.173 µmol/min/mg protein, respectively (Table 8). The activity in methomyl, lambda-cyhalothrin and ∝-cypermethrin- treated strain was almost similar to that in the Lab- strain. As for the SBW larvae, biochemical assays indicated that chlorpyrifos- treated strains, expressed higher levels of AChE activity than the reference (Lab- strain), in which AChE activity was 9.609 µmol/min/mg protein higher than that from the Lab- strain, whereas larvae from profenofos showed somewhat an increase in AChE activity by 4.174 µmol/min/mg protin higher than that in Lab- strain. The activity in methomyl, lambda-cyhalothrin and ∝-cypermethrin-treated strain was almost similar to that in the Lab- strain. Our toxicological data manifestation agreed somewhat with results of field experimental.

Possible alterations of the AChE were recorded, resulted in a reduction in sensitivity to inhibition by chlorpyrifos and profenofos in PBW larvae. The results revealed that synthetic pyrethroids and carbamates resistance was found at a low to medium level in bollworms population which could be related to the reduction in their use in this area (Ishtiaq *et al*. 2012). AChE is a key enzyme in the insect nervous system that hydrolyzes acetylcholine neurotransmitters to terminate nerve impulses, and it is the primary target of OP and carbamate insecticides. Nevertheless, insects have developed resistance to OPs and carbamates through modification of their AChE sensitivity to insecticides (Weill *et al*. 2003, Hemingway *et al*. 2004, and Russell *et al*. 2004). Insensitive AChE as a resistance mechanism to organophosphates and carbamates has been reported in numerous insect species [Voss *et al*., 1980, Fournier and Mutero, 1994, Zhao *et al*. 1996]. The altered AChE would explain the insensitivity of this enzyme to organophosphorus insecticides observed in the field strain.

AChE may have variants with reduced sensitivity to inhibition. Such variants, which may be rare in the wild, unexposed population, may be selected and cause resistance in insects. The insensitive AChE in resistant strains sometimes, but not always, shows a reduced activity to substrates. Because this type of resistance mechanism is caused by a slower rate of reaction with cholinesterase, its effect can be greatly increased by a concomitant augmented detoxication. The amount and activity of AChE are almost always much higher than strictly necessary (Tan *et al*. 2008).

The obtained results are in agreement with those found by Abo Elghar (2005) who stated that AChE activity expressed higher levels in field strains than that of the susceptible strain. Insensitive AChE was identified as a resistance mechanism by comparing biochemical analysis with a laboratory selected monocrotophos resistant cotton bollworm (RR: 200) and the susceptible strain (Ren *et al*. 2002). Generally, AChE from the field strain exhibited a higher insensitivity to the organophosphorus insecticides than the carbamate insecticides. This result is in disagreement with that found by (Yu *et al*. 2003), who stated that AChE from the field strain exhibited a higher insensitivity to the carbamate insecticides than the organophosphorus insecticides.

In regard to the activity of GST in PBW larvae, Table (8) showed that, relatively higher activity of GST has been observed in chlorpyrifos and profenophos-treated strains, over that of the susceptible strain, representing 17.723 and 15.273 mol/min/mg protein respectively. The activity in methomyl-treated strain was almost similar to that in the Lab- strain, whereas larvae from lambda-cyhalothrin and ∝-cypermethrin- treated strains showed somewhat an increase in GST activity higher than that in Lab- strain. The same trend of data was noticed for SPW larvae.

Table (3): The average number and percent reduction of cotton aphid, *Aphis gossypii* per 100 cotton leaves during 2010 and 2011 seasons

Insecticides	Rate of application	Seasons	Pre-treatment	Average No. of Aphid						General mean	% Reduction						General mean	Average of two seasons
				1st spray		2nd spray		3rd spray			1st spray		2d spray		3rd spray		%	
				1 week	2 week	1 week	2 week	1 week	2 week		1 week	2 week	1 week	2 week	1 week	2 week		
α-cypermethrin	250 Cm^3/F	2010	1005	200	260	298	295	315	378	291	80.7	78.3	76.6	77.9	79.0	77.2	78.3	78.1 [a]
		2011	810	150	184	250	240	230	284	187	81.3	79.1	77.8	74.9	78.6	76.1	77.9	
Lambda-cyhalothrin	750 Cm^3/F	2010	970	178	230	268	315	385	450	304.33	82.2	80.1	78.2	75.5	73.4	71.9	76.9	74.1 [b]
		2011	690	150	183	265	254	295	352	249.33	78.05	75.6	72.3	68.8	67.8	65.2	71.3	
Profenophos	750 Cm^3/F	2010	890	150	225	285	320	342	415	289.5	83.6	79.2	74.8	72.9	74.3	71.7	76.1	74.8 [b]
		2011	930	168	208	292	310	385	530	315.5	81.7	79.4	77.4	71.7	68.9	61.1	73.4	
Chlorpyrifos	1000 Cm^3/F	2010	915	180	238	278	325	384	475	313.33	80.9	78.14	76.1	73.3	71.9	68.5	74.8	73.4 [b]
		2011	760	158	184	275	270	335	390	268.66	79.0	77.7	73.9	69.9	66.9	64.9	72.0	
Methomyl	300gm/F	2010	1090	250	325	365	440	545	635	426.66	77.7	74.9	73.6	69.6	76.5	64.7	71.0	68.9 [c]
		2011	730	150	192	280	345	415	484	312.50	79.3	75.8	72.4	59.9	57.3	54.8	66.6	
Untreated		2010	1340	1380	1594	1700	1780	2000	2210	1777.33	–	–	–	–	–	–	–	–
		2011	1030	1020	1120	1430	1215	1370	1510	1277.5	–	–	–	–	–	–	–	–

Values followed by different letters are significantly different.

Table (4): Average number and percent reduction of whitefly, (*Bemisia tabaci*) Immature stage/100 cotton leaves during 2010 and 2011 seasons

Insecticides	Rate of application	Seasons	Pre-treatment	Average No. of whitefly immature stage						General mean	% Reduction						General mean	Average of two seasons
				1st spray		2nd spray		3rd spray			1st spray		2nd spray		3rd spray		%	
				1 week	2 week	1 week	2 week	1 week	2 week		1 week	2 week	1 week	2 week	1 week	2 week		
α-cypermethrin	250 Cm^3/F	2010	254	110	158	170	145	150	175	151.33	49.8	42.1	48.7	55.2	59.7	57.4	52.2	47.1 [a]
		2011	237	135	170	157	175	195	210	173.66	47.5	37.7	45.0	41.9	39.5	37.9	41.9	
lambda-cyhalothrin	750 Cm^3/F	2010	230	120	158	158	165	210	220	171.83	39.5	362	45.0	42.4	39.8	40.8	40.6	39.7 [b]
		2011	250	160	178	178	190	190	220	186	41.0	382	36.9	34.3	44.1	38.3	38.8	
Profenophos	750 Cm^3/F	2010	280	140	175	230	240	253	260	216.83	42.1	41.8	37.1	38.6	40.4	42.6	40.43	39.5 [b]
		2011	344	200	235	255	275	300	330	265.83	46.4	40.7	38.5	37.1	35.8	32.8	38.6	
Chlorpyrifos	1000 Cm^3/F	2010	190	110	130	145	135	160	175	142.5	32.9	36.3	41.5	49.1	44.4	43.1	41.2	40.5 [b]
		2011	330	180	210	250	285	300	315	252.16	36.8	30.7	41.9	38.2	40.0	40.9	39.8	
Methomyl	300 gm/F	2010	256	150	180	190	225	235	242	203.66	32.1	34.5	43.1	37.1	39.4	41.5	37.9	38.2 [b]
		2011	320	200	220	232	250	275	305	247	32.4	40.3	39.8	38.6	36.8	33.2	38.5	
Untreated		2010	285	306	306	306	372	432	460	353.66	–	–	–	–	–	–	–	–
		2011	342	371	394	394	412	412	488	420.66	–	–	–	–	–	–	–	

Table (5): The average number and percent reduction of whitefly, (*Bemisia tabaci*) mature stage /100 cotton leaves during 2010 and 2011 seasons

Insecticides	Rate of application	Seasons	Pre-treatment	Average No. of whitefly mature stage						General mean	% Reduction						General mean	Average of two seasons
				1st spray		2nd spray		3rd spray			1st spray		2nd spray		3rd spray		%	
				1 week	2 week	1 week	2 week	1 week	2 week		1 week	2 week	1 week	2 week	1 week	2 week		
α-cypermethrin	250 Cm^3/F	2010	278	130	143	160	180	198	125	156	44.8	39.5	50.3	43.2	35.3	39.1	42.03	40.04 [a]
		2011	233	130	160	185	200	208	190	178.83	39.9	43.6	40.2	34.1	37.4	33.1	38.06	
lambda-cyhalothrin	750 Cm^3/F	2010	230	120	110	140	160	145	160	139.16	38.5	43.9	47.4	39.03	42.6	32.7	40.7	41.7[a]
		2011	211	100	140	160	175	154	170	149.83	48.9	45.6	42.9	36.4	48.8	33.9	42.8	
Profenophos	750 Cm^3/F	2010	245	115	100	145	125	95	116	116	44.6	52.2	48.9	55.3	64.7	56.6	53.7	54.7 [a]
		2011	229	95	110	125	140	165	120	125.83	55.3	60.6	58.9	53.1	49.4	570	55.7	
Chlorpyrifos	1000 Cm^3 /F	2010	222	117	100	120	135	142	150	127.33	37.8	47.2	53.3	46.7	41.8	34.6	43.6	44.5 [a]
		2011	205	125	135	160	142	120	133	135.83	34.3	45.9	41.2	45.7	58.9	46.8	45.5	
Methomyl	300gm/F	2010	199	109	99	115	135	118	120	116	35.7	41.7	50.1	40.5	46.02	41.7	42.6	43.3 [a]
		2011	184	105	125	131	150	130	120	129.83	38.5	44.3	46.4	37.4	50.4	46.5	43.9	
Untreated		2010	355	301	303	411	405	390	367	362.83	–	–	–	–	–	–	–	–
		2011	320	297	390	425	417	456	390	395.83	–	–	–	–	–	–	–	–

Values followed by different letters are significantly different.

Table (6): Average number and percent reduction of cotton Jassids, *Empoasca* spp./100 cotton leaves during 2010 and 2011 seasons

Insecticides	Rate of application	Seasons	Pre-treatment	Average No. of jassid						General mean	% Reduction						General mean	Average of two seasons
				1st spray		2nd spray		3rd spray			1st spray		2nd spray		3rd spray		%	
				1 week	2 week	1 week	2 week	1 week	2 week		1 week	2 week	1 week	2 week	1 week	2 week		
α-cypermethrin	250 Cm^3/F	2010	102	24	38	32	35	40	32	33.5	69.8	54.2	59.1	49.5	41.2	50.9	54.1	54.7 [c]
		2011	111	23	35	42	38	32	40	35	73.4	55.1	50.3	57.5	55.5	40.5	55.4	
Lambda-cyhaloth rin	750 Cm^3/F	2010	99	31	34	30	22	34	40	31.83	59.8	57.8	60.7	67.3	48.5	36.9	55.2	55.4 [bc]
		2011	115	33	37	35	32	37	40	35.66	60.7	54.2	60.01	65.5	50.4	42.5	55.6	
Profenophos	750 Cm^3/F	2010	105	25	35	29	32	30	39	31.66	69.5	59.02	63.9	55.2	57.1	41.9	57.8	56.6 [b]
		2011	133	34	40	48	56	32	43	42.16	64.9	57.2	52.6	47.7	52.9	46.6	55.3	
Chlorpyrifos	1000 Cm^3 /F	2010	117	35	40	36	40	30	34	35.83	61.6	57.9	59.6	49.7	61.5	54.6	57.5	59.9 [a]
		2011	136	34	40	36	40	35	36	36.83	67.9	63.8	65.5	56.8	61.4	58.6	62.5	
Methomyl	300gm/F	2010	104	39	46	40	38	32	29	37.33	51.9	45.6	49.8	46.3	53.8	56.4	50.6	52.5 [d]
		2011	116	33	39	45	30	36	35	36.33	61.01	52.2	49.1	61.9	53.4	48.7	54.4	
Untreated		2010	150	117	122	115	102	100	96	108.66	–	–	–	–	–	–	–	–
		2011	185	135	130	141	149	120	112	131.16	–	–	–	–	–	–	–	–

Values followed by different letters are significantly different.

Table (7): Average number of total predators and percent reduction / 20 cotton plants during 2010 and 2011 seasons

Insecticides	Rate of application	Seasons	Pre-treatment	Average No. of total predators						General mean	% Reduction						General mean	Average of two seasons
				1st spray		2nd spray		3rd spray			1st spray		2nd spray		3rd spray		N %	
				1 week	2 week	1 week	2 week	1 week	2 week		1 week	2 week	1 week	2 week	1 week	2 week		
α-cypermethrin	250 Cm^3/F	2010	34	12	17	15	19	13	23	16.5	7 2.1	66.7	70.6	65.7	79.8	73.2	71.4	73.5 [a]
		2011	43	13	15	10	13	15	18	14.0	74.7	70.3	79.4	69.8	80.7	79.3	75.7	
Lambda-cyhaloth rine	750 Cm^3/F	2010	39	15	14	12	17	20	22	16.60	69.5	76.1	79.5	73.3	72.9	77.7	74.8	73.3 [a]
		2011	45	13	16	19	13	18	24	17.16	75.8	69.7	62.6	71.1	77.8	73.6	71.8	
Profenophos	750 Cm^3/F	2010	31	11	13	9	17	16	26	15.33	71.9	72.0	76.3	66.4	72.8	66.8	71.03	70.5 [a]
		2011	40	16	14	12	10	23	27	17.0	66.5	70.2	73.5	75.0	68.1	66.6	69.9	
Chlorpyrifos	1000 Cm^3/F	2010	33	14	18	16	14	21	25	18.0	64.2	63.4	67.7	73.9	66.4	70.0	67.6	63.6 [a]
		2011	49	18	20	26	28	31	37	26.66	69.3	65.2	53.1	42.8	64.9	62.6	59.6	
Methomyl	300gm/F	2010	35	19	18	20	16	22	28	20.5	57.2	65.7	61.9	50.0	66.8	68.3	61.7	60.2 [a]
		2011	46	19	24	26	22	30	36	26.16	65.4	51.8	57.7	52.2	63.9	61.3	58.7	
Untreated		2010	38	48	57	57	62	72	96	65.33	–	–	–	–	–	–	–	–
		2011	46	55	54	52	46	83	93	63.83	–	–	–	–	–	–	–	–

Values followed by different letters are significantly different.

Relatively higher activity of GST has been observed in the field populations over that of the susceptible strain in whole larval homogenates (Singh, 2002; Mohan and Gujar, 2003). This result is in agreement with our toxicological data and confirms that GST increased detoxification is involved in the observed resistance to chlorpyrifos and profenofos. Resistance to OPs is considered to be due to metabolism of these compounds by GST. Three major detoxifying enzymes are associated with insecticide resistance: (a) cytochrome P450 monooxygenases, (b) glutathione-*S*- transferases (GSTs), (c) esterases (Bull, 1981; Oppenoorth, 1985). At least one of these stated enzymes in insects is involved in detoxification of insecticides. Enhanced detoxification is usually involved in the evolution of resistance. GSTs are a family of enzymes that catalyze the conjunction of glutathione with electrophilic substrates such as insecticides (Zhao *et al*. 1996). GSTs can metabolize foreign compounds by facilitating their reductive dehydrochlorination or by conjugation reactions with reduced glutathione, to produce water-soluble metabolites that are more readily excreted. In addition, they contribute to the removal of toxic oxygen free radicals produced through the action of pesticides (Vanhaelen *et al*. 2004).

In insects, studies suggest that GSTs play an important role in resistance against several classes of insecticides including OPs and pyrethroids (Wei *et al*. 2001; Hemingway *et al*. 2004). In cases that have been studied in more details, resistance has been attributed to increases in the amount of one or more GST enzymes, either as a result of gene amplification or more commonly through increases in transcriptional rate, rather than qualitative changes in individual enzymes (Grant and Hammock, 1992; Ranson *et al*. 2001).

Table (8) showed that the concentration of total protein in pink and spiny bollworms larvae has been increased in the field populations than of the susceptible strain. Proteins are among most important compound of insects that bind with foreign compounds. The increase in the total protein of treated larvae may reflect the increase in the activity of various enzymes related to organophosphorous, carbamates and pyrethroids. The obtained results are in good agreement with those obtained by Gunning *et al*. 1996 and 1997, who clearly demonstrated that, greater resistance to fenvalerate was accompanied by not only greater enzyme activity but also protein.

Table 8: AChE and GST specific activity in pink and spiny bollworms larvae of laboratory and field strains

Strain	total protein(mg /ml)		AChE(μ mol/min/mg protin		GST(μmol/min/mg protin	
	pink bollworm larvae	spiny bollworm larvae	pink bollworm larvae	spiny bollworm larvae	pink bollworm larvae	spiny bollworm larvae
laboratory strain	1.850 cd	1.520 d	2.803 e	2.760 c	3.130 d	2.980 d
α-cypermethrin*	2.883 bc	3.923 c	3.490 cd	3.298 c	7.198 c	5.770 c
Lambda-cyhalothrin	3.438 ab	4.035 c	3.158 de	3.100 c	6.968 c	5.765 c
Profenophos*	4.130 a	6.320 b	5.173 b	4.174 b	15.273 b	6.377 b
Chlorpyrifos*	2.730 bc	8.713 a	5.897 a	9.609 a	17.723 a	11.163 a
Methomyl*	2.673 bc	4.520 c	3.715 c	3.333 c	4.473 d	3.120 d
Control	1.697 d	1.873 d	3.320 cde	3.387 c	4.773 d	3.450 d

Values followed by different letters are significantly different. * Pesticide-treated strain

4. Conclusion

In light of the aforementioned results, it could be concluded that, the two tested synthetic pyrethroids can be used to control bollworms in cotton fields, followed by methomyl and the two organophosphorus compounds. Despite ∝-cypermethrin, lambda-cyhalothrin and profenophos reduced the number of aphid, they had negative effects on the numbers of the associated natural enemies. The results of biochemical studies revealed that the two synthetic pyrethroids and methomyl were found at a low to medium resistant level in bollworms population which may be due to the reduction in their usage in this area. In general, data emphasized that the repeated application of any insecticide during the same season must be avoided in order to obviate the build up of the resistance phenomenon or secondary pest outbreaks

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